



Forensic Chemistry Standard Operating Procedure Manual Tetrahydrocannabinol Quantitation Procedure for Plant material using GC/MS

36.0 TETRAHYDROCANNABINOL FULL QUANTITATION PROCEDURE FOR PLANT MATERIAL USING GC/MS

36.1 Application

This procedure is used to quantitate the amount of total tetrahydrocannabinol (THC) in plant material samples with GC/MS through a systematic extraction and comparison to a prepared calibration curve of known standard THC concentrations.

36.2 Exhibit Requirements for Analysis

- 36.2.1 Full quantitation will only be performed if requested and approved by laboratory management. Documentation of this approval will be provided in the case file.
- 36.2.2 Cystolithic hairs must be microscopically observed.
- 36.2.3 If cystolithic hairs cannot be detected due to a highly ground plant material sample, a Dugenois-Levine test may serve as a second test.

36.3 Equipment and Reagents

36.3.1 Equipment

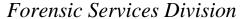
- Gas Chromatograph Mass Spectrometer
- Oven
- Analytical Balance
- Disposable glass tubes with screw caps
- Disposable transfer pipettes
- Disposable autosample vials, inserts, and caps
- Vortexer
- Grinding apparatus
- Assorted volumetric glassware
- Volumetric pipettes with disposable tips

36.3.2 Reagents

- Methanol
- Tribenzylamine
- Δ^9 -Tetrahydrocannabinol two different lot numbers (1 mg/mL in methanol)
- Control spice (suitable non-cannabis plant material)

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36.4 Testing preparation and procedures

36.4.1 Internal standard preparation (300 µg/mL Tribenzylamine)

Weigh 30 mg of tribenzylamine on the analytical balance and dissolve into 100 mL of Methanol using a volumetric flask. Other equivalent ratios may be used to prepare a 300 μ g/mL tribenzylamine solution.

36.4.2 Calibration standard preparation (THC)

The amounts of each reagent and standard to be pipetted are listed in the following table. Each standard will be made in a disposable glass tube and vortexed or shaken until the solution is homogenous.

Calibration standard	Internal standard	Methanol	THC standard (1mg/mL)
50 μg/mL (0.1%)	500 μL	450 μL	50 μL
100 μg/mL (0.2%)	500 μL	400 µL	100 μL
150 μg/mL (0.3%)	500 μL	350 µL	150 µL
300 μg/mL (0.6%)	500 μL	200 μL	300 µL
500 μg/mL (1.0%)	500 μL	-	500 μL

36.4.3 Positive Control and Negative Controls

36.4.3.1 Positive Control preparation (150 µg/mL THC)

Weigh approximately 100 mg of the ground control spice, and record this weight to two decimal places. Pipette 300 μ L of 1 mg/mL THC standard onto this material. This THC standard will be a different lot number than the THC standard used to make the calibration standards when possible.

36.4.3.2 Negative Control preparation

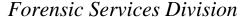
Ground control spice with no added THC will serve as a negative control to ensure no cross contamination of the control spice. Weigh approximately 100 mg of the ground control spice, and record this weight to two decimal places.

36.4.3.3 Pipette 1 mL of 300 µg/mL internal standard solution and 1 mL of methanol onto each control and extract the samples as outlined in 36.4.4

36.4.4 Sample preparation

- 1. Exhibits should have a beginning net weight of 1.0 gram or more for quantitation and sample preservation as discussed in section 11.8. If less than 1.0 gram is available, the analyst will consult their supervisor or the customer on how to proceed.
- 2. After collecting a representative sample for analysis, grind the plant material until a relatively homogenous mixture is formed.

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- 3. Weigh approximately 100 mg of plant material on an analytical balance. Record this weight to two decimal places for percent THC calculations after instrumental analysis.
- 4. Pipette 1 mL of 300 μg/mL internal standard solution and 1 mL of methanol onto the plant material.
- 5. The sample will be vortexed for at least ten minutes.
- 6. The plant material must remain in the extraction solution for a minimum total of 30 minutes before analysis.
- 36.4.5 Obtaining a dry weight of samples for quantitation
 - 1. Any exhibit results that have a total THC percentage of 0.3% or less will require additional quantitation with dry weight samples.
 - 2. Previously ground samples will be dried at minimum for four hours or overnight.
 - 3. After allowing samples to cool to room temperature, perform quantitation procedure.

36.5 Instrumentation

- 36.5.1 A five-point calibration curve using the prepared calibration standards will be generated using the data analysis software for the GC/MS.
- 36.5.2 The GC/MS data will be collected with the appropriate validated method.
- 36.5.3 If exhibits from more than one case will be run in the same sequence, then the sequence file will serve as the unique identifier for that batch of cases. The sequence file will have a uniquely identifiable file name and will be maintained within the FCU.

36.6 Quality Assurance

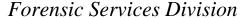
- 36.6.1 Methanol blanks will be run between all calibration standards and samples. These blanks must be free of tribenzylamine and THC for an acceptable autosample run.
- 36.6.2 Clean or disposable glassware will be used in all sample and standard preparations. Disposable pipette tips will be used when pipetting and discarded after use.

36.7 Performance Verification and Acceptance Criteria

- 36.7.1 The calibration points and control standard must be within twenty percent of their expected values to be accepted.
- 36.7.2 If the calibration standards fall outside of the accepted range, the analyst should consider performing instrument maintenance, using a different instrument, and/or remaking the standards.

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36.8 Interpretation

- 36.8.1 The concentration of prepared samples will be calculated using the data analysis software on the GC/MS.
- 36.8.2 The following formula will be used to determine the percent of THC present in the sample.

$$THC \% = \frac{\text{Calculated Concentration } \mu\text{g/mL} \times 100}{(\frac{\text{Weight of Plant material } \mu\text{g}}{2 \text{ mL}})}$$

36.8.3 THC mass spectra must be evaluated for the correct fragmentation pattern.

36.9 Measurement of Uncertainty

- 36.9.1 Measurement of uncertainty will be calculated annually, and all calculations will be maintained in Ensur. Refer to these documents to determine the current measurement of uncertainty.
- 36.9.2 Measurement of uncertainty will be reported when the uncertainty would cause the reported result to be above or below 0.3%.

36.10 Reporting

36.10.1 Samples that have greater than 1% total THC will be reported as follows.

Example 1:

EXHIBIT(S):

001-a Plant material

RESULTS:Controlled SubstanceTotal THC PercentageAmount001-adelta-9-tetrahydrocannabinolGreater than 1%152.20 grams

36.10.2 Only samples with results that are bracketed by the calibration standards will have their actual percentage of total THC reported to two decimal places. Measurement of uncertainty reporting is also demonstrated below.

Example 2:

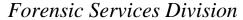
EXHIBIT(S):

001-a Plant material

RESULTS:Controlled SubstanceTotal THC PercentageAmount001-adelta-9-tetrahydrocannabinol0.41%152.20 grams

The measurement has an uncertainty of 0.13% with a 95.4% confidence level, 0.41% ± 0.13%.

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36.10.3 Any exhibits that are below 0.1% total THC will be reported as follows.

Example 3:

EXHIBIT(S):

001-a Plant material

RESULTS:Controlled SubstanceTotal THC PercentageAmount001-adelta-9-tetrahydrocannabinolLess than 0.1%152.20 grams

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